REMARKS

A check for the fee for a three-month extension of time accompanies this response. Any additional fees that may be due in connection with filing this paper or with this application during its entire pendency may be charged to Deposit Account No. 06-1050. If a Petition for extension of time is required, this paper is to be considered such Petition, and any fee charged to Deposit Account No. 06-1050. A change of address for the undersigned accompanies this response.

Claims 1-32, 34-47, 59, 61-64 and 141-147 are pending in this application. Claims 33, 48-57 and 65-83 are cancelled herein without prejudice or disclaimer. Applicant reserves the right to file continuation and divisional applications directed to any cancelled or unclaimed subject matter. Claims 1, 2, 9-11, 32 and 64 are amended herein. Claims 1, 2, and 9-11 are amended to clarify the subject matter of the claims and to address such issues raised in the Office Action. Basis for these amendments can be found throughout the specification, with particular basis, for example, at pages 3, lines 9-25; and page 23, line 29 to page 24, line 2. Claim 32 is amended and Claim 33 is cancelled to remove redundancies with Claim 31.

In the interest of advancing prosecution of the above-captioned application, Claims 9, 10 and 64 are amended herein to delete "*in vivo*." Applicant reserves the right to file continuation or divisional application(s) directed to the omitted subject matter at a future date. Basis for these amendments can be found in the specification, for example, at pages 24-25, describing *in vitro* and *ex vivo* methods.

The Amendment and response, filed September 22, 2003, responsive to the previous Office Action is incorporated in its entirety by reference herein. The Examiner's attention is drawn to the Supplemental Information Disclosure Statement filed on the same day herewith under separate cover.

REJECTION OF CLAIMS 1-57, 59, 61-83 AND 144-147 UNDER 35 U.S.C. §112, FIRST PARAGRAPH

Scope of Enablement

Claims 1-57, 59, 61-83 and 144-147 are rejected under 35 U.S.C. §112, first paragraph, as lacking enablement for the full scope of the claimed subject matter. The Office Action alleges that, although the specification is enabling for methods for introducing a nucleic acid into a cell *in vitro*, it does not provide enablement for *ex vivo* or *in vivo* gene transfer. The Office Action further alleges that because the claims are generic and encompass numerous applications in medical, pharmaceutical, biological and technology related fields, including *in vitro*, *in vivo* and *ex vivo* delivery methods, they necessarily encompass gene therapy applications. The Office Action thus alleges that the specification must enable introducing a nucleic acid into a cell for purposes of gene therapy.

The Office Action further alleges that *ex vivo* and *in vivo* gene therapy is unpredictable. The Office Action alleges that *ex vivo* and *in vivo* gene therapy have generally failed to produce useful results. It is alleged that a myriad of obstacles have been encountered by those of skill in the art seeking to express recombinant proteins in mammals at pharmaceutically relevant levels. It also is alleged that although particular aspects of *ex vivo* and *in vivo* gene therapy were routine at the time of filing, therapeutic application of gene transfer methods remained far from routine. The Office Action concludes that it would not have been possible to practice the instant claimed methods, to the extent that they broadly encompass *ex vivo* and *in vivo* gene therapy, without engaging in undue experimentation

This rejection is respectfully traversed. It is respectfully submitted that in the interest of advancing prosecution, the claims as amended herein do not specify *in vivo* delivery, and thus the rejection as it pertains to *in vivo* delivery has been rendered moot. This rejection also is rendered moot with respect to claims 33, 48-57 and 65-83, which are cancelled herein. Applicant reserves the right to file continuation or divisional application(s) directed to the omitted or cancelled subject matter.

Relevant law

To satisfy the enablement requirement of 35 U.S.C §112, first paragraph, the specification must teach one of skill in the art to make and use the invention without undue experimentation. Atlas Powder Co. v. E.I. DuPont de Nemours, 750 F.2d 1569, 224 USPQ

409 (1984). This requirement can be met by providing sufficient disclosure, either through illustrative examples or terminology, to teach one of skill in the art how to make and how to use the claimed subject matter without undue experimentation. This clause does not require "a specific example of everything within the scope of a broad claim." In re Anderson, 176 USPQ 331, at 333 (CCPA 1973), emphasis in original. Rather, the requirements of §112, first paragraph "can be fulfilled by the use of illustrative examples or by broad terminology." In re Marzocci et al., 469 USPQ 367 (CCPA 1971)(emphasis added).

Further, because "it is manifestly impracticable for an applicant who discloses a generic invention to give an example of every species falling within it, or even to name every such species, it is sufficient if the disclosure teaches those skilled in the art what the invention is and how to practice it." In re Grimme, Keil and Schmitz, 124 USPQ 449, 502 (CCPA 1960). Thus, there is no doubt that a patentee's invention may be broader than the particular embodiment shown in the specification. A patentee not only is entitled to narrow claims particularly directed to the preferred embodiment, but also to broad claims that define the invention without a reference to specific instrumentalities. Smith v. Snow, 294 U.S. 1, 11, 24 USPQ 26, 30 (1935).

Thus, there is no requirement for disclosure of every species within a genus. Applicant is entitled to claims are commensurate in scope not only with what applicant has specifically exemplified, but commensurate in scope with that which one of skill in the art could obtain by virtue of that which the applicant has disclosed.

The inquiry with respect to scope of enablement under 35 U.S.C. §112, first paragraph, is whether it would require undue experimentation to make and use the subject matter *as claimed.* A considerable amount of experimentation is permissible, particularly if it is routine experimentation. The amount of experimentation that is permissible depends upon a number of factors, which include: the quantity of experimentation necessary, the amount of direction or guidance presented, the presence or absence of working examples, the nature of the invention, the state of the prior art, the relative skill of those in the art, the predictability of the art, and the breadth of the claims. Ex parte Forman, 230 USPQ 546 (Bd. Pat. App. & Int'f 1986); see also In re Wands, 8 USPQ2d 1400 (Fed. Cir. 1988).

Analysis

Summary of Arguments

1. The instant claims are directed to methods of introducing large nucleic acid molecules into cells. Dependent claims specify the sizes of the nucleic acid molecules, the types of delivery agents, the types of cells, the order in which the steps of the method are performed, and whether the cells are contacted with the nucleic acid / delivery agent *in vitro* (*e.g.*, cells in culture) or *ex vivo* (cells taken out of the body of a subject). There is no rule in the U.S. patent law that requires enablement of what someone who practices a claimed process or uses a claimed product will do next.

The claims are directed to methods for introducing large nucleic acids into cells either *in vitro* or *ex vivo* (i.e., introduced into cells that have been removed from a subject). The claims do not include any steps that involved gene therapy; and there is no allegation that it requires undue experimentation to harvest cells from a subject. The claims are methods for introducing a certain type of nucleic acid into a cell; they are not directed to methods of gene therapy. This is not to say that someone who practices these methods of introduction and then goes on to use the cells for gene therapy will not infringe the claims; it means that instant claims are not directed to methods of gene therapy and the focus of the scope of enablement should not look beyond the claims.

Hence, the specification is enabling for the full scope of the methods, including the steps of contacting a large nucleic acid with a delivery agent, applying a delivery agent to a cell, and contacting the cell with the large nucleic acid molecule. The specification also is enabling for taking a cell out of the body of a subject, treating the cell with a nucleic acid molecule and a delivery agent(s) *ex vivo*. . As discussed above, the instant claims are not directed to ex vivo gene therapy, but to introduction of nucleic acids into cells that have been removed from a subject.

Contrary to the Examiner's assertion, enablement of the full scope of the instantly claimed methods of nucleic acid delivery into cells does not turn on enablement of uses of the product of the claimed methods, such as gene therapy or the production of desired proteins in transgenic animals. As discussed by Applicant in great detail in the Amendment filed responsive to the previous Office Action and below, and as acknowledged by the Examiner, the specification in light of the knowledge of those of skill in the art is adequately enabling

for the full scope of the methods as claimed, including contacting a large nucleic acid with a delivery agent, applying a delivery agent to a cell, contacting the cell with the large nucleic acid molecule, taking a cell out of the body of a subject, treating the cell with a nucleic acid molecule and a delivery agent(s) *ex vivo*. The claims do not require re-introduction of the cells into a subject nor expression of anything in a subject. Therefore, it is respectfully submitted that the rejection on grounds of inadequate scope of enablement should be withdrawn.

2. It is respectfully submitted that the Examiner is applying an improper standard for enablement by asserting that the instant methods for delivery of nucleic acids into cells are not enabled for their full because a single use, gene therapy, of a product of the instantly claimed methods (a cell containing a heterologous large nucleic acid), was allegedly "unpredictable" as of the application's filing date. Applicant is required to demonstrate that the specification teaches one of skill in the art how to practice the methods as claimed, without undue experimentation. Applicant is not required to enable every use of the product of the claimed methods. Because the specification is enabling for the full scope of the claimed methods of introducing a nucleic acid molecule into a cell by treating the cell *in vitro* or *ex vivo*, it is respectfully submitted that the claims are enabled for their full scope, regardless of the state of the art of gene therapy.

3. Notwithstanding the impropriety of the rejection for inadequate scope of enablement on grounds of the alleged unpredictability of one use, gene therapy, of the product of the claimed methods, it is respectfully submitted that the Examiner has failed to establish that the state of the art of gene therapy was "unpredictable" as of the application's filing date. The Examiner's alleges that gene therapy is unpredictable because every embodiment in every disease condition for every animal model system was not demonstrated as of the application's filing date.

Enablement does not require demonstration of every embodiment of a method, nor of a fully perfected, optimized method. The references cited by the Examiner and provided by the Applicant responsive to the previous and instant Office Actions clearly demonstrate that gene therapy was successfully implemented as of the application's filing date, and the references further provide detailed descriptions of parameters that can be tested and / or modified to make continued advancements in the field. Applicant clearly has established that

at the time the instant application was filed, it would have required no more than routine experimentation to follow the teachings of the successes of gene therapy and apply them to new disease states or animal models. The amount of experimentation required to use the products of the instantly claimed methods in gene therapy, was not undue. Therefore, regardless of the impropriety of the rejection on grounds of inadequate scope of enablement because of the alleged unpredictability of the use of a product of the instantly claimed methods, and regardless of the further impropriety of an allegation of such unpredictability based on lack of demonstration of gene therapy as applied to every disease condition and in every animal model system, it is respectfully submitted that the art of gene therapy as of the instant application's filing was not so unpredictable as to render the instant methods of delivering large nucleic acid molecules into cells inadequately enabled for their full scope.

4. A consideration of the factors enumerated in In re Wands, even taking into account the state of the art of *ex vivo* gene therapy as a possible application of the instant claims, leads to the conclusion that the specification in light of the knowledge of those of skill in the art is adequately enabling for the full scope of the methods as claimed.

1. A determination of enablement or scope of enablement turns on whether the methods *as claimed* are enabled for their full scope

The instantly claimed methods are directed to introducing a large nucleic acid molecule into a cell *in vitro* or *ex vivo* . The claims specify the order in which the large nucleic acid molecule, the cell and a delivery agent are contacted with each other, and whether particular delivery agents are used in the methods. The methods include the steps of contacting a nucleic acid molecule with a delivery agent, applying an agent to a cell and contacting the nucleic acid molecule with the cell. The claims also include the use of energy, including ultrasound and electroporation, with a delivery agent to introduce nucleic acid molecules into cells. The claims further include contacting the nucleic acid molecule with the cell *in vitro*, *e.g.*, with a cell in culture, or *ex vivo*, with a cell that is taken out of the body of a subject.

Enablement requires that the application teach one of skill in the art how to practice the methods *as claimed*, without undue experimentation. As demonstrated in the analysis below (*see* Section 4), a consideration of the factors enumerated in In re Wands leads to the conclusion that any experimentation necessary to practice the methods as claimed is routine,

and its amount is not undue. The application teaches one of skill in the art to make and use the full scope of the claimed methods of introducing a nucleic acid molecule into a cell, whether the cell is in culture *in vitro* or is taken out of the body of a subject and treated *ex vivo*.

Enablement and, likewise, scope of enablement, should turn on whether a nucleic acid molecule can be delivered to a cell by following the steps of the method as claimed. As is discussed more fully in the In re Wands analysis below, the application enables one of skill in the art to make and use the claimed subject matter. The application teaches one of skill in the art how to perform the methods as claimed to introduce a nucleic acid molecule into a cell. The specification provides numerous working examples and descriptions of methods of delivering nucleic acids such as: introduction of a nucleic acid molecule using specific ordered steps; the use of energy and cationic compounds as delivery agents; the use of combinations of delivery agents; introduction of large nucleic acid molecules; and the introduction of nucleic acids into a variety of cell types. The application further describes how to recognize the products of the method. For example, the application describes methods of labeling, detecting labeled nucleic acid molecules and measuring the delivery of nucleic acid molecules. Such methods for detecting the product of the methods are exemplified in the working examples.

Further, unless a claim is limited by a use recited in the claim, a determination of enablement or lack thereof should not be premised on a particular use. As set forth in the MPEP 2164.01(c), "if any use is enabled when multiple uses are disclosed, the application is enabling for the claimed invention." Applicant has met the burden of demonstrating that one of skill in the art can make and use the subject matter as claimed. The claims are directed to introducing a nucleic acid into a cell. They are not limited by a particular use for such cell. The specification provides multiple uses for cells with an introduced nucleic acid molecule in medical, pharmaceutical, biological and technology-related research fields including production of proteins, generation of transgenic animals, uses with imaging methods for medical devices, mapping biological events, gene delivery optimization technology development, and therapies. Enablement requires only that one of these uses be enabled.

In addition, the Office Action acknowledges that the application is enabled for methods of introducing a nucleic acid into a cell. For example, the Office Action states that

the application is enabled introducing a nucleic acid into a cell *in vitro* (sentence spanning pages 3-4). The Office Action further acknowledges that methods of *ex vivo* delivery were routine at the time of filing including transfection of cells *in vitro*, followed by selection and introduction of cells into a subject (paragraph spanning pages 7 and 8). Therefore, it is respectfully submitted that Applicant has enabled the methods *as claimed*, and the rejection should be obviated.

2. Applicant is not required to enable uses of *the products* of the claimed methods

Enablement of the claimed methods does not require the Applicant to enable every use of the product of the instantly claimed methods. The Office Action challenges the scope of enablement based on whether therapeutic application of gene transfer methods are enabled. For example, the Office Action states "the skilled artisan would not be able to make and use the full scope of the invention at least insofar as it encompasses methods of *ex vivo* and *in vivo* gene therapy or the production of pharmaceuticals in animals." Further, it is alleged that "it would not have been possible to practice the instant claimed method to the extent it broadly encompasses *ex vivo* and *in vivo* gene therapy without engaging in undue experimentation because achieving therapeutic effect by gene transfer was far from routine at the time of filing."

The claims are not directed to methods of gene therapy, but to methods of introducing nucleic acid into cells either *in vitro* or *ex vivo*. Specifying that the introduction is *ex vivo* refers to the source of the cells. It does not require reintroduction of the cells into a subject. The reintroduction of the cells is a subsequent step that is not part of the claimed method.

Therapeutic effects and therapies are *uses of the product* of the methods as claimed that have no bearing on whether by following the steps of the method as claimed, one can obtain the desired result, in this case, introduction of a large nucleic acid molecule into a cell. The claims are directed to introducing a nucleic acid molecule into a cell. The claims encompass contacting a cell, such as a cell in culture (*in vitro*) or a cell removed from a subject (*ex vivo*), to a delivery agent and a nucleic acid molecule in a manner that introduces the nucleic acid molecule into the cell. Although the cells containing the nucleic acid molecule produced by the method can be used in further steps such as replacement into a

subject or for subsequent therapies, these are only some uses of a cell containing a large nucleic acid molecule; Applicant is not required to enable every one of these uses.

It is respectfully submitted that it is not incumbent upon Applicant to teach one of skill in the art how to use *the product* of the claimed methods. Enablement requires only that one of skill in the art can make and use *the claimed method.* As an analogy, if one were to claim "a method of hitting a golf ball by striking the ball, whereby the ball moves forward," one would only be required to demonstrate that the method was enabled for striking the ball and moving the ball forward. Whether the golf ball moved forward onto a green, into a hole, into a lake or sand trap would not be relevant to an analysis of enablement of the method as claimed.

In the claims at issue, although one of skill in the art can use the claimed methods to obtain a cell containing a nucleic acid and then use that product (*i.e.*, a cell containing a large nucleic acid that is introduced into the cell) in subsequent methods or uses, the claims are directed to methods of obtaining the product. Although the specification describes a wide variety of uses for cells containing introduced nucleic acid molecules, these are uses of *the product of the claimed methods,* not how to practice the claimed methods. In the golf ball analogy, where the ball travels after it is hit by the method is not relevant to enablement of a method for striking a golf ball. Similarly, enablement of the use of a cell after introducing a nucleic acid into the cell has no bearing on enablement of the methods of introducing large nucleic acid molecules into cells as instantly claimed. Applicant respectfully submits that the instant methods are enabled, regardless of whether gene therapy is enabled. As discussed below, however, regardless of the impropriety of the determination of inadequate scope of enablement of the instant methods based on an alleged lack of enablement of gene therapy, it is respectfully submitted that the art of gene therapy, including *in vitro* and *ex vivo* therapy, was enabled at the time the instant application was filed.

The Examiner appears to suggest that Applicant's statements made responsive to the previous Office Action are at cross purposes because Applicant allegedly states, on the one hand, that "all of the instant claims are directed to methods for delivering nucleic acid molecules into cells, not methods of gene therapy," then allegedly acknowledges, "the uses of nucleic acid delivery, including gene transfer for gene therapy, constitute only a subset of the embodiments that are within the scope of the subject matter as instantly claimed." In

response, it is respectfully submitted that it is not contradictory to state that *one use* of the product of the claimed methods of nucleic acid delivery into cells, is gene therapy. Furthermore, the reference in the previous response to the uses of the cells for gene therapy are closer to embodiments in which the claims specified *in vivo* introduction of the nucleic acid molecule. Resolution of the issues regarding introduction of nucleic acids into cells *in vivo* has been rendered moot in this application.

The Examiner, on the other hand, appears to premise scope of enablement of the instant methods of nucleic acid delivery on the alleged lack of enablement of a *single use* of the product of the claimed methods, gene therapy, rather than on the methods as claimed. It is respectfully submitted that not only does the Examiner improperly base his conclusion of inadequate scope of enablement of the instant methods on the alleged unpredictability of an *application* of the methods, gene therapy, but the Examiner's conclusion of inadequate scope of enablement is premised on a consideration of *only one factor* of the In re Wands analysis, predictability, and predictability of an application rather than predictability of the methods *per se*, out of the several that must be considered in a determination of whether it would require undue experimentation to practice the claimed methods. As discussed previously and below, regardless of the impropriety of a rejection of inadequate scope of enablement of a method based on alleged lack of enablement of a single use of that method, the Examiner has failed to demonstrate that the state of the art of gene therapy was so unpredictable as of this application's filing date that a consideration of the factors enumerated in In re Wands would lead to a determination that it would require undue experimentation to practice the instantly claimed methods.

3. Gene Therapy, including *in vitro* and *ex vivo* therapy, was not so unpredictable as of the application's filing date to render the instant claims inadequately enabled for their full scope.

As discussed above, to demonstrate the full scope of enablement of the claimed methods, Applicant is not required to demonstrate enablement of gene therapy using cells containing an introduced nucleic acid molecule (the products of the instantly claimed methods), since the instant claims are not directed to methods involving gene therapy. Further, even if scope of enablement of the instantly claimed methods turned on the enablement of gene therapy, it is respectfully submitted that a consideration of the factors

enumerated in In re Wands leads to the conclusion that it would require no more than routine experimentation, and the amount of such experimentation would not be undue, to use the instant methods to carry out gene therapy, including *in vitro* and *ex vivo* therapy, as an application of the instant claims.

Applicant respectfully submits that methods of *in vitro* and *ex vivo* therapy, such as can be performed with cells produced by the instantly claimed methods, were enabled at the time of filing. *Ex vivo* therapy involves introducing a nucleic acid molecule into a cell after removing cells from the body of a subject. The specification demonstrates that using the claimed methods, the nucleic acid molecules can be introduced into cells, whether the cells are cultured *in vitro* or removed from a subject for *ex vivo* introduction, and that the introduced nucleic acid molecules are intact and functional for gene expression after introduction into cells (*see*, Examples 4-7). Applicant knows of no reason, and the Examiner provides no evidence, which would indicate that a method acknowledged by the Examiner as being enabled for introduction of a large nucleic acid molecule into a cell in culture *in vitro*, is not enabled for *ex vivo* introduction of a large nucleic acid molecule into a cell that has been removed from the body of a subject.

The specification describes and exemplifies introduction of nucleic acid molecules into cells in a wide variety of cell types and with numerous embodiments of the methods. The specification also describes methods for introducing such cells into a subject. Further, Applicant has provided numerous references available at the time of filing that demonstrate a wide variety of nucleic acid molecules introduced into cells *in vivo* and *ex vivo* are functional (*see*, *e.g.*, references submitted in the Supplemental Disclosure Statement filed September 22, 2003 and discussed in the previous Response filed September 22, 2003). Further supporting references are provided herein (*see*, references submitted herewith and made of record in a supplement Information Disclosure Statement submitted under separate cover: Isner *et al.*(1999) *J. Clin. Invest. 103(9)*: 1231-1236; Springer *et al.* (1998) *Molecular Cell 2*:549-558; Yoo *et al.* (2000) *Clin. Orthop. 379 Suppl.*: S164-70; Asahara *et al.* (2000) *Gene Therapy 7*:451-57). These references demonstrate that introduced nucleic acid molecules are expressed *in vivo* and *ex vivo* in a wide variety of cell types. The references further demonstrate expression and function of introduced nucleic acids including marker genes as well as genes for *in vivo* and *ex vivo* therapies. The In re Wands analysis below further

discusses the state of the art of *ex vivo* therapy at the time of filing in relation to the claims at issue.

The Examiner alleges that in addressing the art cited by the Examiner in the previous Office Action as allegedly supportive of the unpredictability of gene therapy, Applicant points to "statements indicating enthusiasm for the future of gene therapy" as evidence of enablement of the methods. The Examiner states that "statements that the methods will one day be enabled" are not evidence that the method was routinely available at the time of filing. Applicant respectfully disagrees. In discussing each of the references pointed to by the Examiner, Applicant showed how the references teach successes in gene therapy, pointed to limitations cited in the references that guide the direction for continued experimentation, and demonstrated that any experimentation required to apply gene therapy to other systems is at best routine, given the teachings of these references and the advanced knowledge of those of skill in the art. None of the references cited by the Examiner point to lack of enablement and/or a mere hope of future enablement. To the contrary, as discussed extensively by Applicant in the previous response, the references cited by the Examiner as being indicative of lack of enablement in fact teach the parameters that are important to achieve success in gene therapy in other systems, which in turn makes further experimentation by one of skill in the art to achieve additional successes in gene therapy, all the more routine. The Examiner is equating a recognition of the need for growth and advancement in the field of gene therapy as an indication that the state of the art of gene therapy was not yet "enabled." As discussed below, the standard for enablement is not a fully optimized method or product. Rather, it is whether, given the knowledge of those of skill in the art, there is sufficient disclosure, through demonstrative examples and other description, to adapt a method such as gene therapy to a system of interest with no more than routine experimentation.

The cited references do not simply make enthusiastic statements that gene therapy will work sometime in the future. Rather, they teach the successes of gene therapy, provide specific direction and guidance for future experimentation and improvements, and state that the field will continue to grow and improve. The Examiner is applying an improper standard for enablement by equating a recognition of improvements that can be made in a field such as gene therapy, with lack of enablement of the state of the art of gene therapy. In effect, the

Examiner appears to be stating that no science is enabled unless it is completely perfected, with no further need for advances or improvements.

Responsive to Applicant's assertion in the previous response that the Examiner appears to equate limitations with unpredictability, the Examiner rebuts that given these limitations, one of skill in the art would be able to predict "immediate failure" in attempting to practice the "majority" of the embodiments encompassed by the claimed method. The Examiner then rebuts that while the references cited in the previous Office Action may achieve expression of a given gene *in vitro* or in an animal system, this "clearly cannot be taken as evidence" for claims that broadly encompass gene therapy of *any* condition and in systems other than certain animal models.

It again is respectfully submitted that the standard applied by the Examiner for enablement of the instant methods is improper. Not only does the Examiner incorrectly assert that in order that the steps of the instant methods be enabled, one of its potential applications, gene therapy, must be enabled, but the Examiner further asserts that for gene therapy to be enabled, *every* disease condition in *every* animal model system must have been successfully treated by gene therapy at the time the instant application was filed. The Examiner is in effect stating that the proper standard of enablement of a particular method is that (1) every one of the *uses of its products* must be enabled; and (2) a use of a product of a method is only enabled if *every single embodiment* of the use has been carried out with success as of the application's filing date. Such a fully optimized, perfected method is clearly not the standard for enablement, and the Examiner is further elevating that standard by applying it to every embodiment of *applications* of a claimed method. Rather, **a considerable amount of experimentation is permissible**, particularly if it is routine experimentation.

Far from predicting "immediate failure," the references cited in the previous Office Action teach parameters that provide guidance on how to apply gene therapy to systems other than those provided by the references. As discussed in great detail in the previous Office Action, the references provide examples of successful gene transfer clinical trials, state how more such trials are necessary, provide specific parameters, such as gene delivery methods and gene expression control systems, that can be modified depending on the particular system, and further provide recommendations on how to vary such parameters or conduct

such studies. The cited references teach the pitfalls to be avoided in future experiments, limitations to be taken into account, and how to overcome these limitations. For example, Orkin *et al.* and Marshall *et al.* teach the limitations of viral vectors in gene delivery, then suggest that these limitations may be overcome by the use of cationic lipids as gene delivery agents – far from being limitations that predict "immediate failure," these are limitations for which the cited references provide solutions, and some of these solutions, *e.g.*, choice of cationic lipids as delivery agents and the development of artificial chromosomes as vectors, are in fact adopted by the methods as instantly claimed.

The "enthusiastic statements" made by the cited references are based on demonstrations of specific and numerous successes of gene therapy, including human gene transfer and clinical trials (*e.g.*, Eck *et al.* and Orkin *et al.)* as discussed extensively by Applicant in the previous response. The successes described in these references, along with the provision of parameters that govern modification of the technique for a different embodiment of gene therapy, predict more such successes along with continuing advancements in the field. The prediction of future successes based on current successes would not be possible if the experimentation required to apply gene therapy to disease conditions or animal models other than the specific examples provided in the references were not routine or "predictable." If, as the Examiner alleges, the references cited in the previous Office Action provide no guidance on how to apply their teachings to further experimentation in gene therapy, and the references only teach limitations in gene therapy that are harbingers of "immediate failure," then there would be no basis for their statements that urge continued funding of gene therapy research (*e.g.*, Orkin *et al.*) or predict dramatic improvements in the technology (*e.g.*, Marshall *et al.*) or specify particular parameters, such as gene delivery methods and types of studies that have been implemented with success and/or that could be modified in the future to overcome any existing limitations (*e.g.*, Rubanyi *et al.*). Clearly, given the teachings of the cited references, one of skill in the art is apprised of the feasibility of gene therapy, is aware of particular limitations, and knows how to predictably and systematically address these limitations without undue experimentation. Even if the experimentation is considerable, it is routine, given the teachings of the references and the advanced knowledge of those of skill in the art.

The additional references cited by the Examiner in the instant Office Action as allegedly demonstrating the "unpredictability" of gene therapy or of the *in vivo* production of proteins of interest, provide no deviation from the teachings of the references cited by the Examiner in the previous Office Action, or provided by Applicant in response thereto, regarding the predictability of the state of the art of gene therapy and the requirement of no more than routine experimentation to modify its application, depending on the particular disease. The Examiner cites two references, Somia *et al.* (*Nature Rev. Genet.*, 1:91-99 (2000)) and Rosen *et al.* (*New Engl. J. Med.*, 346:1185-1193 (2002)) that allegedly contradict the teachings of Cavazzana-Calvo *et al.*, cited by Applicant responsive to the previous Office Action. It is alleged that these references state that the successful treatment of severe combined immunodeficiency (SCID) taught by Cavazzana-Calvo *et al.* is due to this gene being particularly suited as a target for therapy. Although these references discuss the suitability of the SCID gene as a target for gene therapy, neither makes any statement that teaches the unpredictability of gene therapy. Rosen is directed to the treatment of SCID, and like Cavazzana-Calvo *et al.*, demonstrates the success of gene therapy. Rosen makes a single statement at the end that the methods applied to gene therapy of SCID may not be as easily applicable in the case of genes whose expression must be tightly regulated. Rosen does not teach that no other examples of the success of gene therapy exist, or that gene therapy is not enabled or predictable. Somia *et al.* describes the success of treating SCID, and provides detailed parameters for the construction of vectors that can help to overcome safety concerns with gene therapy. Applicant's extensive discussion of references cited by the Examiner and by Applicant in the previous and instant Office Actions clearly demonstrates the many successes of gene therapy as of the instant application's filing date, and the knowledge regarding specific modifications for advancement or improvement of the art of gene therapy. It is therefore respectfully submitted that the state of the art of gene therapy, combined with the knowledge of those of skill in the art, clearly demonstrates that any modifications by one of skill in the art to apply gene therapy to a disease state of interest would require no more than routine experimentation. Enablement of gene therapy does not require that it be fully optimized or successfully demonstrated in every single embodiment / disease state. Rather, it is whether gene therapy could be practiced without undue experimentation at the time the instant application was filed, and the Examiner has provided no evidence to the contrary.

The Examiner cites yet another reference, Houdebine (*Transgenic Res.*, 9:305-320, (2000)), for the proposition that the production of recombinant proteins in mammals at "pharmaceutically relevant levels," is unpredictable. Applicant respectfully submits that, like gene therapy, the generation of transgenic animals has no bearing on the enablement of the instant methods of introducing large nucleic acid molecules into cells, where the cells are treated *in vitro* or *ex vivo* after removal from the body of a subject. Applicant is not required to enable every possible use of the product of the instantly claimed methods, namely, a cell containing a large nucleic acid molecule that is introduced into the cell. The irrelevance notwithstanding, Houdebine does teach the generation of transgenic animals using transfected somatic cells as nuclear donor, and the expression of transgenes therein. Houdebine teaches that the transgene expression is "predictable to a limited extent," and has yet to be optimized. Houdebine does not teach that the expression of transgenes in transgenic animals is unpredictable – to the contrary, the reference discusses the successes of transgene expression at an industrial scale, *e.g.*, in the production of proteins in milk and in the egg white of transgenic chickens, and the generation of transgenic farm animals, albeit at high cost. As discussed above, the standard for enablement is not a completely perfected or optimized technology, nor of a technology that has been demonstrated in every possible embodiment. Houdebine teaches how to generate transgenic animals and obtain transgene expression in a "predictable" fashion, and even makes a statement to that effect (*see* Abstract).

Therefore, regardless of the irrelevance of transgenic animal technology to a method for introducing a nucleic acid into a cell as instantly claimed, the Examiner has provided no evidence that transgenic animals and transgene expression could not be practiced at the time the instant application was filed. Again, it is respectfully submitted that the Examiner is equating a fully optimized procedure, successfully demonstrated in every embodiment, with enablement.

In conclusion, it is respectfully reiterated that the Examiner has incorrectly applied enablement of *every embodiment* of *every use* of the *product* of the instantly claimed methods as the standard for enablement of the methods as instantly claimed. The impropriety of the rejection notwithstanding, it is further submitted that although the art of gene therapy and of transgene expression may not have been fully optimized and perfected at the time the instant application was filed, it was not so unpredictable as to qualify as a major factor in the

determination of whether the requirements of 35 U.S.C. § 112, first paragraph, are satisfied with respect to the instantly claimed subject matter.

As discussed below, the instant claims are directed to methods of introducing a nucleic acid molecule into a cell. In some embodiments, the cells are contacted with the nucleic acid molecule *in vitro* or *ex vivo*. As the Examiner has acknowledged, the specification is enabling for introducing a large nucleic acid molecule *in vitro* or *ex vivo*, using the combinations of delivery agents and order of steps as instantly claimed. *Ex vivo* contact of a cell with a nucleic acid molecule means that the cell is taken out of the body of a subject, then contacted with a nucleic acid molecule. As the Examiner has acknowledged, the specification is enabling for *ex vivo* introduction of a nucleic acid molecule into a cell, then introducing the cell back into the subject. Therefore, it is respectfully submitted that the specification is enabling for the methods as claimed. Further, although the claims are not directed to gene therapy methods, the disclosure in the specification, in light of the knowledge of those of skill in the art and the predictability of gene therapy as discussed above, is enabling for *ex vivo* therapy applications of the products of the instant methods.

4. Analysis of enablement: Application of In re Wands factors to the methods as instantly claimed

Scope of the claims

Claims 1-32, 34-48, 59, 61-64, and 144-147 are directed to methods for introducing a nucleic acid molecule into a cell. Some claims (*e.g.*, Claims 9, 10, 64) specify steps of contacting the nucleic acid with the delivery agent and/or the cell *in vitro* or *ex vivo*.

Independent Claim 1 is directed to a method of introducing a large nucleic acid molecule into a cell by (a) contacting a large nucleic acid molecule with a delivery agent, (b) applying a delivery agent to a cell, and (c) contacting the cell with the nucleic acid molecule. Steps (a) and (b) are performed sequentially in any order, followed by step (c), except if the delivery agent is energy, it is not applied to the nucleic acid molecule or to the cell after contacting the cell with the nucleic acid molecule. Independent Claim 34 is directed to a method of delivering a nucleic acid molecule to a cell where the cell is contacted with a delivery agent and ultrasound or electrical energy and then the cell is contacted with the nucleic acid molecule. Independent Claim 59 is directed to a method for delivering a large nucleic acid molecule into a cell that includes (a) contacting the nucleic acid molecule with a

composition that comprises a cationic lipid composition of DOSPA and DOPE, where the nucleic molecule is at least 5 megabases in size; and then (b) contacting the nucleic acid molecule with a cell, wherein steps (a) and (b) are performed simultaneously or sequentially. Claims that depend on the independent claims recited above specify elements including size ranges and types of nucleic acid molecules; types of delivery agents; treatments and parameters for using the delivery agents; the order in which the steps of the methods are practiced; and cell types. Additionally, dependent Claims 9 and 10 (dependent on Claim 1) and Claim 64 (dependent on Claim 59) specify methods of delivering nucleic acid molecules *in vitro*, and *ex vivo*.

Level of Skill

The level of skill in this art is recognized to be high (see, *e.g.*, Ex parte Forman, 230 USPQ 546 (Bd. Pat. App. & Int'f 1986)). The numerous articles and patents made of record in this application address a highly skilled audience and further evidence the high level of skill in this art.

Teachings of the specification

As discussed in great detail in the Amendment filed responsive to the previous Office Action, the specification teaches methods of delivery of nucleic acid molecules into cells. The specification provides numerous descriptions of methods of delivering nucleic acids such as: introduction of a nucleic acid molecule using specific ordered steps; the use of energy and cationic compounds as delivery agents; the use of combinations of delivery agents; introduction of large nucleic acid molecules; and the introduction of nucleic acids into a variety of cell types. Additionally, as acknowledged by the Examiner on pages 3-4 of the instant Office Action, and as made of record by Applicant responsive to the previous Office Action, the application teaches methods of introducing a nucleic acid into a cell *in vitro* (*see, e.g.*, page 23, line 28 to page 24, line 30).

The specification further describes that nucleic acid molecules can be delivered to primary cell lines, such as fibroblast, muscle, stomach, intestine, lung, bladder, ovary, uterus, liver, kidney, pancreas, brain, heart, spleen, prostate to mimic *in vivo* systems (page 26, lines 5-8).

Knowledge of those of skill in the art

As discussed, introduction of cells into a subject is not part of the instantly claims methods. The instant claims are directed to introduction of large nucleic acid molecule into cells; there is no step of subsequent introduction into a subject.

Notwithstanding this, as acknowledged in the Office Action and as made of record by Applicant responsive to the previous Office Action, a broad body of knowledge existed in the art related to methods and reagents introduction of DNA into cells at the time of filing. Additionally, methods for introducing cells into a subject for *ex vivo* delivery were also known in the art, including the introduction of cells for a wide variety of diseases, conditions and tissue types. Many methods were available in the art for introduction of a wide variety of cell types into tissues and organs. Exemplary references are described further below and provided herein: Isner *et al.*(1999) *J. Clin. Invest. 103(9)*: 1231-1236; Springer *et al.* (1998) *Molecular Cell 2*:549-558; Yoo *et al.* (2000) *Clin. Orthop. 379 Suppl.*: S164-70; Asahara *et al.* (2000) *Gene Therapy 7*:451-57. These references are made of record in the accompanying Information Disclosure Statement.

Presence of working examples

The specification provides numerous working examples and descriptions of methods of delivering nucleic acids such as: introduction of a nucleic acid molecule using specific ordered steps; the use of energy and cationic compounds as delivery agents; the use of combinations of delivery agents; introduction of large nucleic acid molecules; and the introduction of nucleic acids into a variety of cell types. The specification also describes methods of labeling, detecting labeled nucleic acid molecules and measuring the delivery of nucleic acid molecules. Additionally, methods of gene expression and detecting gene expression from nucleic acid molecules delivered to cells are described. The specification teaches that *ex vivo* delivery of a nucleic acid molecule can be accomplished by introducing a nucleic acid molecule into a cell and then introducing such cell into a subject. Thus, the examples are applicable for delivery of a nucleic acid to any cell for *in vitro* and *ex vivo* delivery.

Delivery of nucleic acids to a broad variety of cell types and with many embodiments of the methods is exemplified in Examples 4-7. Example 4 describes the introduction of ACes into Chinese Hamster lung fibroblast cells using the delivery agent Superfect and then

contacting the Superfect:ACes complex with the cells (page 47, lines 1-20 and page 47, line 28 through page 48, line 4). Examples 5 and 6 describe the introduction of GFP chromosomes using ultrasound and Saint-2 or ultrasound and Lipofectamine (pages 50-52) into a variety of cell types such as CHO-KI, Hep-G2, A9 and V79-4 cells. Tables 1 and 2 of Example 7 (pages 54-57) exemplify the use of numerous transfection protocols using delivery agents such as clonfectin, cytofectene, Eu-fectins, lipofectamine and superfect. Measurement of gene expression from delivered nucleic acids is exemplified in Example 1 (page 41, lines 1-25), Example 5 (page 48, line 15 through page 49, line 31). Example 6 (page 50, line 1 through page 51, line 4) and Example 7 (page 54, lines 4-6). Detection of the delivery of nucleic acids by detecting labeled cells is exemplified in Example 4 (page 47, line 22 through page 48, line 4) and Example 7 (page 55, line 8 through page 56, line 5 and also Table 2, pages 56-57).

Predictability

The instantly claimed methods are directed to methods of delivering a nucleic acid molecule to a cell. The level of knowledge and skill in the delivery of nucleic acids into cells as claimed in the instant application was high as of the effective filing date. The Office Action acknowledges that the specification teaches introducing a nucleic acid molecule into a cell *in vitro* as claimed (paragraph spanning pages 3-4). The Office Action further acknowledges that transfection of cells *in vitro*, followed by selection and introduction of cells into a subject, were routine at the time of filing and are taught by the specification (paragraph spanning pages 7-8). Introduction of cells *ex vivo* similarly is taught by the specification, since the only difference between *in vitro* and *ex vivo* is the source of the cells into which the nucleic acid is introduced.

Furthermore, the Office Action also acknowledges that *ex vivo* delivery, which encompasses delivery of a nucleic acid molecule to a cell, followed by introduction of the cell into a subject, is taught by the specification and was routine at the time of filing.

Therefore, given the extensive teachings of the specification, in combination with what was known at the time the instant application was filed, it is not unpredictable that nucleic acid molecules, including large nucleic acid molecules can be delivered to cells according to the steps of the instant methods.

Although the claims are not directed to gene therapy, the delivery of nucleic acid molecules for *ex vivo* gene therapy also is not unpredictable. First, *ex vivo* delivery involves introduction of a nucleic acid molecule into a cell *in vitro*, where the cell is taken from the body of a subject. As taught by the specification and as acknowledged in the Office Action, it is not unpredictable to be able to deliver nucleic acid molecules into cells *in vitro* as instantly claimed; therefore, delivery of nucleic acid molecules into cells treated *ex vivo* after being taken from the body of a subject also is predictable. Second, *ex vivo* delivery can involve the introduction of the cells containing the introduced nucleic acid molecule into a subject. The Office Action acknowledges that transfection of cells *in vitro*, followed by selection and introduction of cells into a subject, were routine at the time of filing (paragraph spanning pages 7-8). Additionally, numerous methods for introducing a wide variety of cells into subjects were available in the art as of the filing date of the application. For example, it had been demonstrated in the art that transplanted cells expressed genetically engineered genes *in vivo* following introduction and that the cells successfully integrated into target tissues in the recipient. Exemplary references available at the time of filing demonstrating successful *ex vivo* delivery are described below: Isner *et al.*(1999) *J. Clin. Invest. 103(9)*: 1231-1236; Springer *et al.* (1998) *Molecular Cell 2*:549-558; Yoo *et al.* (2000) *Clin. Orthop. 379 Suppl.*: S164-70; Asahara *et al.* (2000) *Gene Therapy 7*:451-57.

Isner *et al.* describes the transfer of bone marrow cells. Cells expressing a *lacZ* marker gene under the control of an endothelial cell promoter were transplanted into recipient mice. Subsequent induced injuries, such as corneal injury, demonstrated that the implanted cells successfully migrated to the site of injury, incorporated into corneal neovascularized tissue and expressed the marker gene (see pages 1234-35 and Figure 1, page 1233). Isner *et al.* also details cell therapy with enthothelial progenitor cells (EPCs) including cells engineered to express proangiogenic factors (Figure 3, page 1234). In animal models for ischemia, EPCs were shown to incorporate into active sites of neovascularization (page 1234, paragraph 1).

Springer *et al.* describes implantation of myoblasts into muscle. Myoblast cells engineered to express VEGF and a lacZ marker gene were transplanted into mice tibialis anterior and lateral gastrocnemius. Examination of the mice legs at 11-47 days post-transplantation demonstrated that the cells incorporated into complex vascular structures and

expressed VEGF. Further, the implanted tissue gave a physiological response to the VEGF-expressing cells (see for example, page 550, columns 1-2 and page 553, last paragraph, column 2).

Yoo *et al.* describes *ex vivo* therapy by transplanting cells for osteogenic regeneration. Yoo *et al.* further describes successful orthotopic and heterotopic bone formation using progenitor cells and *ex vivo* gene transfer (page S168, column 2, second paragraph). Further, Yoo *et al.* describes successful delivery of bone marrow cells by venous injection and injection into the marrow. Successful transplantation of stromal ells expressing collagen Type I and population of the transplanted cells in bone, marrow, lung, cartilage, and spleen of the recipient is also presented. Additionally, Yoo *et al.* describes an efficient method for engraftment of cells in a localized bone region resulting in proliferation and survival of the cells *in vivo* (page S169, column 1).

Asahara *et al.* describes successful transplantation of stem cells including hematopoietic stem cells (HSCs) endothelial progenitor cells (EPCs), neural stem cells (NSCs) and mesenchymal stem cells (MSCs). Foe example, Asahra *et al.* describes the successful *ex vivo* culturing and transplantation of EPCs, such as cells expressing VEGF, and their incorporation into vascular tissue (page 454, col. 1). Asahara *et al.* describe *ex vivo* therapy with genetically engineered neural stem cells. The genetically engineered cells were differentiated *in vitro* into oligodendrocytes, expanded and then transplanted to a demyelinated area of a recipient animal. The transplanted cells resulted in the remyelination of axons. Asahara *et al.* also describe successful transplantation of NSCs secreting never growth factor leading to the amelioration of the death of striatal projection neurons (page 454, col. 2). Additionally, Asahara *et al.* describes *ex vivo* therapy with MSCs for replacement of circulating proteins and engraftment of murine stromal cells following intravenous infusion (page 454, col. 2).

Conclusion

In light of the extensive teachings and examples in the specification for the delivery of a nucleic acid molecule to a cell, the high level of skill of those in this art, the knowledge of those of skill in the art, the fact that it is predictable given the teachings of the instant application and the state of the art at the time of filing to deliver nucleic acid molecules into a cell and introduce the cell into a subject, including carrying out *ex vivo* gene therapy, it would

not require undue experimentation for one of skill in the art to make and use the method of nucleic acid molecule delivery methods as taught by the instant application for *in vitro* or *ex vivo* delivery. Accordingly, a consideration of the factors enumerated in In re Wands leads to the conclusion that undue experimentation would not be required to introduce a nucleic acid molecule into a cell for delivery *in vitro* or *ex vivo*, using the delivery agents and order of steps of the methods as instantly claimed.

Policy Considerations

A significant portion of the grounds for the rejection of the claims under 35 U.S.C. §112, first paragraph, is based on the alleged unpredictability of using cells containing introduced large nucleic acid molecules, which are products of the instant methods, for gene therapy, such as *ex vivo* therapy. As discussed above, the instant methods are directed to nucleic acid delivery into cells, and this delivery may be carried out by contacting the nucleic acid molecules with *in vitro* cultures of cells, or cells that have been taken out of the body of a subject and treated *ex vivo*. The Office Action acknowledges that the steps of the methods of delivery as claimed are enabled for *in vitro* introduction of nucleic acids into cells. It is respectfully submitted that the steps of introducing a nucleic acid molecule into a cell as instantly claimed are no different whether the cell is in culture *in vitro*, or is taken out of the body of a subject and contacted with a nucleic acid molecule *ex vivo*. The Office Action further acknowledges that reintroduction of cells into a subject after *ex vivo* treatment of the cells was routine as of the application's filing date. Furthermore, as demonstrated extensively by the Applicant previously and herein, although the instant claims do not specify a particular use, such as therapy, of the cells containing an introduced large nucleic acid molecule, methods of *ex vivo* therapy were clearly established as of the application's filing date. Accordingly, Applicant is entitled to claims that are commensurate in scope not only with what Applicant has specifically exemplified, but commensurate in scope with that which one of skill in the art could obtain by virtue of that which the Applicant has disclosed. In the instant application, Applicant provides the public with methods of delivering large nucleic acids into cells. As a broad body of knowledge is available in the area of cell culture and manipulation, whether the cells are cultured *in vitro*, or established as primary cultures after being taken from a subject, or simply taken from a subject and treated *ex vivo*, it would be

unfair, unduly limiting and contrary to the public policy upon which the patent laws are based to require Applicant to limit these claims to introducing a large nucleic acid molecule into a cell *in vitro*. To limit an Applicant to claims involving the specific materials disclosed in the examples so that a competitor, seeking to avoid infringement can merely follow the disclosure and make routine substitutions "is contrary to the purpose for which the patent system exists - to promote progress in the useful arts"). *See, e.g.*, In re Goffe, 542 F.2d 801, 166 USPQ 85 (CCPA 1970).

> The public purpose on which the patent law rests requires the granting of claims commensurate in scope with the invention disclosed. This requires as much the granting of broad claims on broad inventions as it does the granting of more specific claims on more specific inventions" In re Sus and Schafer, 49 CCPA 1301, 306 F.2d 494, 134 USPQ 301, at 304.

To require Applicant to further limit the claims would permit those of skill in the art to practice what is disclosed in the specification but avoid infringing claims so-limited. If Applicant is required to limit the claims to introduction of large nucleic acid molecules into cells *in vitro*, then those of skill in the art could by virtue of the teachings of this application readily practice what is claimed by taking cells from the body of a subject and contacting them with a large nucleic acid molecule *ex vivo* and, although not specified as a step of the instant claims, even reintroduce the cells into the subject, a step that the Office Action acknowledges was routine at the time the instant application was filed. To permit that is simply not fair. The instant application exemplifies the introduction of large nucleic acid molecules into cells *in vitro*. Having done so, it is now routine for others to carry out the steps of the instant methods on cells that have been taken out of the body of a subject and contacted with a large nucleic acid molecule *ex vivo*. Those of skill in the art should not be permitted to make such minor modifications by substitution of the source of the cells used in the instant methods of delivering nucleic acid molecules into cells.

REJECTION OF CLAIMS 1-33 and 144-146 UNDER 35 U.S.C. §112, SECOND PARAGRAPH

Claims 1-33 and 144-146 are rejected under 35 U.S.C. §112, second paragraph as being indefinite because it is alleged that the recitation in Claim 1 of "exposing" is indefinite and that it is unclear what constitutes exposing a nucleic acid molecule or a cell to a delivery agent. It also is alleged that Claim 1 is indefinite in reciting that the steps can be performed

"in any order." Specifically, it is alleged that if step (a) is performed first, it would not be possible to perform step (c) second without performing step (b) simultaneously. Therefore, it is alleged that performing steps in the order of (a), (c), (b) are eliminated within the scope of performing steps (a) -(c) sequentially. Claims 2-33 and 144-146 are allegedly indefinite insofar as they depend upon Claim 1. Reconsideration is respectfully requested in light of the amendments herein and the following remarks.

Claim 1 is amended herein to clarify the language by replacing "exposing" with "contacting" such that step (a) of the method recites "contacting a large nucleic acid molecule with a delivery agent." Claim 1 also is amended such that step (b) recites "contacting a cell with a delivery agent." The amendments provide clarity and consistency with the language in other pending claims, such as independent Claims 34 and 59. Basis for these amendments can be found throughout the specification, with particular basis, for example, at page 3, lines 9-25; and at page 23, line 29 to page 24, line 2.

Claim 1 also is amended herein to recite that steps (a) and (b) are performed sequentially in any order, followed by step (c). Basis for this amendment may be found in the specification, for example, at page 2, lines 10-29; page 3, lines 9-25; and page 18, lines 3-13. Thus, the issue of step (c) preceding step (b) and occurring simultaneously with step (b) is rendered moot.

REJECTION OF CLAIMS 1, 3-10, 12-14 and 30-33 UNDER 35 U.S.C. §102(b)

Claims 1, 3-10, 12-14 and 30-33 are rejected under 35 U.S.C. §102(b) as being anticipated by Hadlaczky *et al.* (U.S. Patent No. 6,025,155). It is alleged that the instant claims are interpreted to encompass the method where steps (b) and (c) are performed simultaneously. Therefore, it is alleged that Hadlaczky *et al.*, which allegedly teaches a method of nucleic acid delivery where a cell is contacted with simultaneously with a nucleic acid molecule and a delivery agent, anticipates the claimed subject matter. Reconsideration of this rejection is respectfully requested in view of the amendments herein and the following remarks. It is respectfully submitted that this rejection has been rendered moot with respect to Claim 33, which has been cancelled.

Relevant law

Anticipation requires the disclosure in a single prior art reference of each element of the claim under consideration. In re Spada, 15 USPQ2d 1655 (Fed. Cir, 1990), In re Bond,

15 USPQ 1566 (Fed. Cir. 1990), Soundscriber Corp. v. U.S., 360 F.2d 954, 148 USPQ 298, 301, adopted 149 USPQ 640 (Ct. Cl.) 1966. See, also, Richardson v. Suzuki Motor Co., 868 F.2d 1226, 1236, 9 USPQ2d 1913,1920 (Fed. Cir.), cert. denied, 110 S.Ct. 154 (1989). "[A]ll limitations in the claims must be found in the reference, since the claims measure the invention." In re Lang, 644 F.2d 856, 862, 209 USPQ 288, 293 (CCPA 1981). Moreover it is incumbent on Examiner to identify wherein each and every facet of the claimed invention is disclosed in the reference. Lindemann Maschinen-fabrik Gmbh v. American Hoist and Derrick Co., 730 F.2d 1452, 221 USPQ 481 (Fed. Cir. 1984). Further, the reference must describe the invention as claimed sufficiently to have placed a person of ordinary skill in the art in possession of the invention. An inherent property has to flow naturally from what is taught in a reference In re Oelrich, 666 F.2d 578, 581, 212 USPQ 323, 326 (CCPA 1981).

"Rejections under 35 U.S.C. §102 are proper only when the claimed subject matter is identically disclosed or described in the "'prior art'" . . .the [r]eference must clearly and unequivocally disclose the claimed compound or direct those skilled in the art to the compound without any need for picking, choosing, and combining various disclosures not directly related to each other by the teachings in the cited references. Such picking and choosing may be entirely proper when making a rejection of a 103, obviousness rejection, where the applicant must be afforded an opportunity to rebut with objective evidence any inference of obviousness which may arise from the similarity of the subject matter which he claims to the prior art, but it has no place in the making of a 102, anticipation rejection." (Emphasis in original). In re Arkey, Eardly, and Long, 455 F.2d 586, 172 USPQ 524 (CCPA, 1972).

THE CLAIMS

Independent Claim 1 is directed to a method of introducing a large nucleic acid molecule into a cell by (a) contacting a large nucleic acid molecule with a delivery agent, (b) applying a delivery agent to a cell, and (c) contacting the cell with the nucleic acid molecule. Steps (a) and (b) are performed sequentially in any order, followed by step (c), except if the delivery agent is energy it is not applied to the nucleic acid molecule or to the cell after contacting the cell with the nucleic acid molecule. Dependent claims further specify sizes of nucleic acid molecules (Claims 3-6); that the nucleic acid molecule is an artificial chromosome (Claim 7); methods of contacting the nucleic acid molecule with the delivery

agent or the cells with the nucleic acid *in vitro*, and *ex vivo* (Claims 9 and 10); cationic compounds as delivery agents (Claims 12-14); and cell types (Claims 30-32). Thus, the rejected claims all are directed to a method of nucleic acid delivery to a cell where the nucleic acid is exposed to a delivery agent, a delivery agent is applied sequentially either before or after the previous step to a cell, and the two preceding steps are followed by contacting the cell with the nucleic acid molecule.

ANALYSIS

The Examiner alleges that Hadlaczky *et al.* anticipates the instant claims because it discloses steps (b) and (c) being performed simultaneously. As discussed above, Claim 1 is amended herein to clarify the claim language to specify that steps (a) and (b), contacting a large nucleic acid molecule with a delivery agent and applying a delivery agent to a cell, are performed sequentially in any order, while step (c), contacting the cell with the nucleic acid molecule, follows steps (a) and (b). Thus, Claim 1 as amended for clarity requires step (c) to follow step (b), and steps (b) and (c) are not performed simultaneously.

Since anticipation requires that a reference disclose every element as claimed, Hadlaczky *et al.*, which does not disclose a method where steps (a) contacting a large nucleic acid molecule with a delivery agent, and (b) applying a delivery agent to a cell, are performed sequentially in any order, followed by step (c) of contacting the cell with the nucleic acid molecule, Hadlaczky *et al.* does not anticipate the instant Claim 1. Since Claims 3-10, 12-14 and 30-32 are dependent on Claim 1, and therefore incorporate all the limitations of Claim1, Hadlaczky *et al.* also does not anticipate any of Claims 3-10, 12-14 and 30-32.

REJECTION OF CLAIMS 1, 2-17, 26-28, 30-33, 61-64, 141-144 and 146 UNDER 35 U.S.C. §103(a)

Claims 1, 2-17, 26-28, 30-33, 61-64, 141-144 and 146 are rejected under 35 U.S.C. §103(a) as being unpatentable over Hadlaczky *et al.* U.S. Patent No. 6,025,155 in light of Unger *et al.* (1997) *Invest. Radiol.* 32:723-727. The Office Action alleges that Hadlaczky *et al.* teaches that artificial chromosomes of 20-30 Mb can be introduced by lipid-mediated transfer. It is further alleged that one of ordinary skill in the art would understand such transfer to include (a) exposing the nucleic acid molecule to a delivery agent, (b) exposing the cell to a delivery agent and (c) contacting the cell with the nucleic acid molecule, where the

steps are performed sequentially in any order. The Examiner concludes that Hadlaczky *et al.* teaches all the limitations of Claims 1 and 3-8, except for embodiments where one of the delivery agents is energy.

Unger *et al.* is alleged to teach a method for introducing a nucleic acid molecule into a cell by (a) exposing the nucleic acid molecule to a delivery agent, (b) exposing the cell to a delivery agent and (c) contacting the cell with the nucleic acid molecule, where in step (a) the nucleic acid is exposed to a cationic compound and in step (b) the cell is exposed to ultrasound. It is specifically alleged that Unger *et al.* teaches a method where energy (ultrasound) is applied 30 minutes before the addition of transfection agent. The Examiner concludes that Unger *et al.* teaches all the limitations of Claims 1, 2, 11 and 26-28, except for a large nucleic acid molecule.

Furthermore, the Office Action alleges that there is motivation to combine the cited references because Hadlaczky *et al.* allegedly teaches that artificial chromosomes are useful for expressing exogenous DNA in a cell and Unger *et al.* allegedly teaches that ultrasound enhances gene expression in all cell types tested. Therefore, one of skill in the art could allegedly combine the teachings of the references to obtain the claimed subject matter. The Office Action additionally alleges that Hadlaczky *et al.* and Unger *et al.* teach the limitations of Claims 9, 10, 12-17, 30-33, and 61-64, as well as provide all of the components for a kit of Claims 141-143. It is alleged that Hadlacsky *et al.* also teaches nucleic acid molecules of the size ranges recited in Claims 144 and 146.

Reconsideration of this rejection is respectfully requested in view of the amendments herein and the following remarks.

Relevant Law

In order to set forth a prima facie case of obviousness under 35 U.S.C. § 103: (1) there must be some teaching, suggestion or incentive supporting the combination of cited references to produce the claimed invention (ACS Hospital Systems, Inc. v. Montefiore Hospital, 732 F.2d 1572, 1577, 221 USPQ 329, 933 (Fed. Cir. 1984)) and (2) the combination of the cited references must actually teach or suggest the claimed invention. Further, that which is within the capabilities of one skilled in the art is not synonymous with that which is obvious. Ex parte Gerlach, 212 USPQ 471 (Bd. APP. 1980). Obviousness is tested by "what the combined teachings of the references would have suggested to those of ordinary skill in

the art." In re Keller, 642 F.2d 413, 425, 208 USPQ 871, 881 (CCPA 1981), but it cannot be established by combining the teachings of the prior art to produce the claimed invention, absent some teaching or suggestion supporting the combination (ACS Hosp. Systems, Inc. v Montefiore Hosp. 732 F.2d 1572, 1577. 221 USPQ 329, 933 (Fed. Cir. 1984)). "To imbue one of ordinary skill in the art with knowledge of the invention in suit, when no prior art reference or references of record convey or suggest that knowledge, is to fall victim to the insidious effect of a hindsight syndrome wherein that which only the inventor taught is used against its teacher" W.L. Gore & Associates, Inc. v. Garlock Inc., 721 F.2d 1540, 1553, 220 USPQ 303, 312-13 (Fed. Cir. 1983).

Under 35 U.S.C. §103, in order to set forth a case of prima facie obviousness, the differences between the teachings in the cited reference must be evaluated in terms of the whole invention, and the prior art must provide a teaching or suggestion to the person of ordinary skill in the art to have made the changes that would produce the claimed product. See, *e.g.*, Lindemann Maschinen-fabrik Gmbh v. American Hoist and Derrick Co., 730 F.2d 1452, 1462, 221 U.S.P.Q.2d 481, 488 (Fed. Cir. 1984). The mere fact that prior art may be modified to produce the claimed product does not make the modification obvious unless the prior art suggests the desirability of the modification. In re Fritch, 23 U.S.P.Q.2d 1780 (Fed. Cir. 1992); see, also, In re Papesh, 315 F.2d 381, 137 U.S.P.Q. 43 (CCPA 1963). In addition, if the proposed modification or combination of the prior art would change the principle of operation of the prior art invention being modified, then the teachings of the references are not sufficient to render the claims prima facie obvious. In re Ratti, 270 F.2d 810, 123 USPQ 349 (CCPA 1959).

THE CLAIMS

Claim 1 and Claims 2-17, 26-28, 30-33, 144 and 146 dependent thereon

Independent Claim 1 is directed to a method of introducing a large nucleic acid molecule into a cell by (a) contacting a large nucleic acid molecule with a delivery agent, (b) contacting the cell with a delivery agent, and (c) contacting the cell with the nucleic acid molecule. Steps (a) and (b) are performed sequentially in any order, followed by step (c), except if the delivery agent is energy it is not applied to the nucleic acid molecule or to the cell after contacting the cell with the nucleic acid molecule. Dependent claims further specify types and properties of delivery agents (Claims 2, 11-17, 26-28), sizes and types of

nucleic acids (Claims 3-8, 144 and 146), methods involving *in vitro* and *ex vivo* contact of the cells with the nucleic acids and/or delivery agents (Claims 9-10), and cell types (Claims 30-33).

Claims 61-64

Claims 61-64 depend upon independent Claim 59. Claim 59 is directed to a method for delivering a large nucleic acid molecule into a cell by (a) contacting the nucleic acid molecule of at least 5 megabases with a composition containing cationic lipids DOSPA and DOPE.; and then (b) contacting the nucleic acid molecule with a cell; where steps (a) and (b) are performed simultaneously or sequentially. Dependent Claim 61 specifies that the DNA is a natural chromosome, an artificial chromosome, a fragment of a chromosome, or naked DNA. Claims 62 and 63 specify particular cell types for use with the method of Claim 59. Claim 62 recites a plant cell and an animal cell. Claims 63 recites primary cell, an immortalized cell, an embryonic cell, a stem cell, a transformed cell and a tumor cell. Claim 64 is directed to the method of Claim 59 where the nucleic acid molecule is contacted with the cell *in vitro* or *ex vivo*.

Claims 141-143

Independent Claim 141 is directed to a kit for delivering nucleic acids into cells containing a composition containing an artificial chromosome, a composition containing a delivery agent, reagents for performing sonoporation or electroporation and optionally, instructions for delivering nucleic acids into cells. Dependent Claims 142 and 143 species articular categories and species of delivery agents.

Differences Between the Claims and the Teachings of the Cited References

Hadlaczky *et al.*

Hadlaczky *et al.* does not teach or suggest methods of delivering DNA to cells that include the particular order of steps of (a) contacting a large nucleic acid molecule with a delivery agent, (b) applying a delivery agent to a cell, sequentially in any order, followed by step (c), contacting the cell with the nucleic acid molecule. Hadlaczky *et al.* does not teach or suggest specific orders of steps for the introduction of nucleic acid molecules into cells. Hadlaczky *et al.* also does not teach or suggest methods of DNA delivery using ultrasound energy. Hadlaczky *et al.* does not teach or suggest methods that include contacting a nucleic acid with a combination of DOSPA and DOPE as a delivery agent. Hadlaczky *et al.* also

does not teach or suggest components of a kit which include combinations of an artificial chromosome, a delivery agent and reagents for sonoporation or electroporation .

Unger *et al.*

Unger *et al.* is directed to delivery of plasmid DNA to cells using ultrasound energy. In the methods of Unger *et al.* plasmid DNA is complexed with lipsomes and applied to cells. Ultrasound energy is applied to the cells after the liposome-DNA complex is applied to the cells, to increase the transfection efficiency. Unger *et al.* does not teach or suggest that when energy is applied to the cells, it is applied before contacting the cells with the nucleic acid molecule. Although Unger *et al.* describes an experiment in which ultrasound energy is applied to the cells 30 minutes before adding a transfection agent, Unger *et al.* teaches that the optimal conditions that facilitate transfection involve exposing the cells to ultrasound energy after adding the liposome-DNA complex to the cells. Therefore Unger et al. teaches away from applying energy to the cells before contacting the cells with the liposome-DNA complexes.

Unger *et al.* does not teach or suggest delivery of large nucleic acid molecules. Unger *et al.* also does not teach or suggest methods of DNA delivery by contacting a large nucleic acid with compositions of DOSPA and DOPE and contacting the nucleic acid with a cell. Unger *et al.* also does not teach or suggest components of a kit that includes combinations of an artificial chromosome, a delivery agent and reagents for sonoporation or electroporation.

ANALYSIS

It is respectfully submitted that the Examiner has failed to set forth a case of *prima facie* obviousness. As detailed below, the combination of teachings of Hadlaczky *et al.* with the teachings of the Unger *et al.* does not result in the instantly claimed methods of delivering a large nucleic acid to a cell. The combination of teachings of Hadlaczky *et al.* with the teachings of the Unger *et al.* also does not result in the instantly claimed kits for delivering a large nucleic acid to a cell.

The combination of teachings of Hadlaczky *et al.* with the teachings of the *Unger et al.* does not result in any of the instantly claimed methods of Claims 1 and Claims 2-17, 26-28, 30-32, 144 and 146 dependent thereon.

Neither Hadlaczky *et al.* nor Unger *et al.*, singly or in combination, teaches or suggests the instantly claimed methods for delivering a large nucleic acid to a cell. The methods as claimed specify the steps of (a) contacting a large nucleic acid molecule with a delivery agent, (b) contacting a cell with a delivery agent , and (c) contacting the cell with the nucleic acid molecule. Steps (a) and (b) are performed sequentially in any order, followed by step (c), except if the delivery agent is energy it is not applied to the nucleic acid molecule or to the cell after contacting the cell with the nucleic acid molecule.

Hadlaczky *et al.* does not teach or suggest a particular order of steps for introducing a large nucleic acid molecule into a cell, nor of applying energy as a delivery agent. Unger *et al.* does not cure these deficiencies. First, Unger *et al.* provides no teaching or suggestion of methods for introducing large nucleic acid molecules into a cell. The methods of Unger *et al.* are limited to plasmid DNAs of less than about 10 kB. Further, Unger *et al.* teaches *against* the instantly claimed subject matter. Unger *et al.* demonstrates that the most efficient introduction of plasmid DNA into cells is achieved by incubating a liposome/DNA complex with cells followed by application of ultrasound to the cell/nucleic acid mixture 60 minutes after transfection. Unger *et al.* demonstrates (*see* Figure 4) that application of ultrasound to cells before adding liposome/DNA complexes results in significantly less efficient transfection than the transfection efficiency when ultrasound is applied after adding liposome/DNA complexes to cells. Thus, Unger *et al.* teaches selection of conditions for introducing nucleic acids into cells in which ultrasound energy is applied to the cells *after* contacting the cells with nucleic acid (lipid-DNA complexes); in fact, Unger *et al.* explicitly teaches selection of these conditions (page 725, col. 1, second full para.). Unger *et al.* teaches the selection of applying ultrasound after contacting the nucleic acid and the cells, and teaches away from applying ultrasound before contacting the nucleic acid with the cells. "A prior art reference must be considered in its entirety, *i.e.*, as a whole, including portions that would lead away from the claimed invention" (emphasis in the original). W.L. Gore & Associates, Inc. v Garlock, Inc., 721 F.2d 1540, 220 USPQ 303 (Fed. Cir. 1983). One of skill in the art, by following the method of Unger *et al.*, would select the application of ultrasound *after* the nucleic acid molecule has been contacted with the cell, not *before* the nucleic acid molecule has been contacted, as explicitly taught by Unger *et al.* Thus, Unger *et al.* teaches away from the claimed subject matter of Claim 1, which specifically recites that if the

delivery agent is energy, it is not applied to the cell after contacting the cell with the nucleic acid molecule.

The Office Action contends that Figure 4 of Unger *et al.* teaches treatment of ultrasound before contacting the cell with the nucleic acid. However, as discussed above, Figure 4 demonstrates that introduction of ultrasound before contacting the cell with the nucleic acid results in less efficient transfection efficiency than applying ultrasound after contacting the cell with the nucleic acid. There is no suggestion to choose a less efficient method. Simply disclosing a less optimal condition does not suggest to one of skill in the art that such conditions should be chosen to introduce a large nucleic acid. To the contrary, describing a method as being less desirable suggests *away* from its selection.

It is respectfully submitted that "obvious to experiment" is not the standard for obviousness. "There must be a reason or suggestion in the art for selecting the procedure used, other than the knowledge learned from the applicants' disclosure." In re Dow, 837 F.2d 469, 473 (Fed. Cir. 1988). Unger *et al.* has not provided any teaching or suggestion for the selection of applying ultrasound before contacting the cell with the nucleic acid; in fact, Unger *et al.* suggests not to select such conditions. Therefore, the combination of the teachings of Hadlaczky *et al.*, which does not teach any application of ultrasound energy as a delivery agent, with the teachings of the Unger *et al.*, which does not teach delivery of any large nucleic acid molecule to a cell and especially not by applying energy to the cell before contacting the cell with the nucleic acid, does not result in the instantly claimed method of Claim 1 or any of Claims 2-17, 26-28, 30-33, 144 and 146, dependent thereon. Therefore, the Examiner has failed to establish a *prima facie* case of obviousness.

The combination of the teachings of Hadlaczky *et al.* with the teachings of the Unger *et al.* does not result in any of the instantly claimed methods of Claims 61 -64.

Claims 61-64 are dependent on Claim 59. Thus, if Claim 59 is not obvious in view of the combined teachings of the cited references, Claims 61-64 dependent thereon are not obvious. Combining the teachings of Hadlaczky *et al.* with the teachings of Unger *et al.* does not result in the method as set forth in Claim 59, directed to a method for delivering a large nucleic acid molecule into a cell by (a) contacting the nucleic acid molecule of at least 5 megabases with a composition containing cationic lipids DOSPA and DOPE.; and then (b) contacting the nucleic acid molecule with a cell, where steps (a) and (b) are performed

simultaneously or sequentially. Hadlaczky *et al.* does not teach or suggest the particular combination of DOSPA and DOPE as a delivery agent for introduction of a large nucleic acid molecule into a cell.

Unger *et al.* does not cure this deficiency. Unger *et al.* is directed to introduction of plasmid DNAs; it does not teach or suggest any methods for introducing large DNA of at least 5 megabases into cells, and it further does not teach or suggest introducing DNA of any size into cells using a combination of DOSPA and DOPE as delivery agents. Unger *et al.* teaches combinations of DPEPC/DOPE to introduce plasmid DNA into cells. There is no teaching or suggestion in Unger *et al.* of any other combination of cationic lipids that can be used for introducing nucleic acids into cells.

Therefore, neither Hadlaczky *et al.* nor Unger *et al.*, singly or in combination, teaches or suggests the combination of delivery agents as claimed herein of DOSPA and DOPE for introducing nucleic acid molecules, particularly nucleic acid molecules of at least 5 megabases, into cells. Unger *et al.* does not teach or suggest that any of the methods disclosed in the reference are applicable to large DNAs. The methods taught by Unger *et al.* pertain to plasmid DNAs of less than about 10 kB. Thus, it is not taught or suggested that the combination of DPEPC/DOPE used by Unger *et al.*, nor any other combinations of cationic lipids, would be applicable to methods of introducing a large nucleic acid molecule of at least 5 megabases into a cell. Therefore, the combination of the teachings of Hadlaczky *et al.* with the teachings of Unger *et al.* does not result in the instantly claimed method of Claim 59. Since Claims 61-64 depend from Claim 59 and incorporate all of the limitations of Claim 59, the combination of the teachings of Hadlaczky *et al.* with the teaching of Unger *et al.* does not result in the instantly claimed methods of Claims 61-64. Therefore, the Examiner has failed to establish a *prima facie* case of obviousness.

The combination of teachings of Hadlaczky *et al.* with the teachings of the *Unger et al.* does not result in any of the instantly claimed kits of Claims 141-143.

Neither Hadlaczky *et al.* nor Unger *et al.*, singly or in combination, teaches or suggests the instantly claimed kits for delivering nucleic acids into cells containing a composition containing an artificial chromosome, a composition containing a delivery agent, reagents for performing sonoporation or electroporation and optionally, instructions for

delivering nucleic acids into cells. Hadlaczky *et al.* does not teach or suggest any kits, much less the components of the instant kit claims that include an artificial chromosome, a delivery agent and reagents for sonoporation or electroporation.

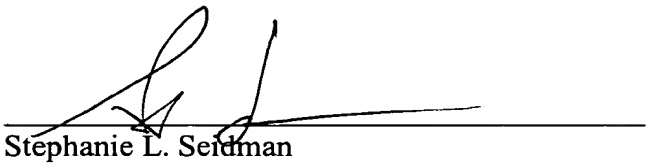
Unger *et al.* does not cure this deficiency. Unger *et al.* also does not teach or suggest any kits. Therefore, the combination of the cited references cannot result in the instantly claimed kits.

The Examiner alleges at page 21 of the Office Action that "the combined teachings of Hadlaczky *et al.* and Unger *et al.* provide all of the components of the kits of Claims 141-143." In response, it is respectfully submitted that neither reference nor their combination teaches or suggests assembly of any components into kits. Further, not only does neither reference teach any kits, but there is no teaching or suggestion in either of the cited references for combining the components of the kits as instantly claimed. Hadlaczky *et al.* does not teach or suggest combining an electroporation or sonoporation reagent with a delivery agent for delivering large nucleic acid molecules. Unger *et al.* also does not provide any teaching or suggestion for combining components such as an artificial chromosome, a delivery agent and a sonoporation or electroporation reagent into a kit. Therefore, neither reference can cure the other's deficiencies because neither reference teaches or suggests selection of the particular combination of components of the instantly claimed kits. As set forth in MPEP 2141.01, "The references must be considered as a whole and must suggest the desirability and thus the obviousness of making the combination." There is no suggestion in Hadlaczky *et al.* or Unger *et al.*, singly or in combination, of the desirability of combining a large nucleic acid molecule such as an artificial chromosome, a delivery agent and a sonoporation or electroporation agent into a kit for delivering artificial chromosomes into cells. The Examiner is improperly engaging in hindsight by picking and choosing elements of each of the references to form components of a kit as instantly claimed. Therefore, it is respectfully submitted that the Examiner has failed to establish a *prima facie* case of obviousness.

* * *

In view of the above amendments and remarks, reconsideration and allowance of the application are respectfully requested.

Respectfully submitted,



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